Standardized Approach for Assessing Potential Risks to Amphibians Exposed to Sediment

NAVFAC
(Naval Facilities Engineering Command)

Presentation Overview Slide

- Project Background and Amphibian Biology
- Project Phases
  - Phase 1
    - Literature review and development of amphibian screening values
  - Phase 2
    - Development of laboratory testing techniques for amphibians exposed to sediment
  - Phase 3
    - Validation of laboratory testing techniques
  - Phase 4
    - Development of guidance manual for assessing potential risks to amphibians at Naval facilities
- Summary and Conclusions

What's the big deal with Amphibians???

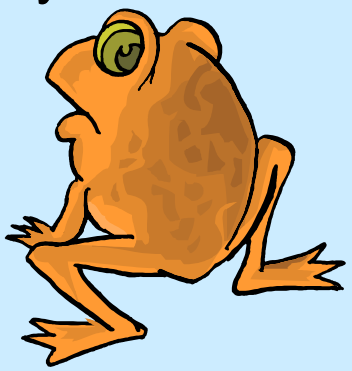
- Wetlands can comprise a substantial portion of open space at many Naval facilities
- Wetlands are prime habitat for amphibians
- Amphibians play a key ecological role serving both as an important prey and predator in wetlands
- Limited amphibian ecotoxicity data are available
- Wetland risk management decisions are often made using inappropriate species (e.g., fathead minnows) that may not be typical of the wetland

What's the big deal with Amphibians??? (cont.)

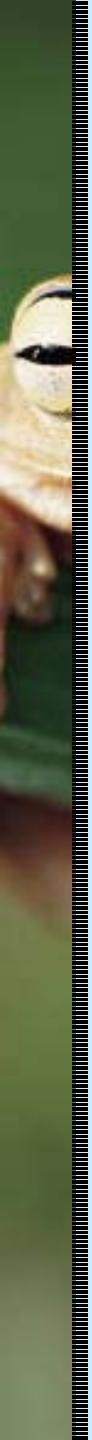
- Global declines documented since the 1980s
- Possible factors include:
  - Changes in atmospheric conditions
  - Habitat loss/alteration
  - Invasive species interactions
  - Exposure to disease and pathogens
  - Chronic and acute exposure to environmental contaminants

Specific Program Objectives

- Selected by the Alternative RemediationTechnology Team (ARTT) to be funded through the Navy's Pollution Abatement Ashore Technology Demonstration/Validation Program – Y0817
- Develop a standardized risk assessment protocol for evaluating potential risks to amphibians at Navy sites
- Protocol can be used to help the Navy avoid costly and unnecessary wetland alteration based on use of inappropriate ecological endpoints



Amphibian Biology



Amphibian Taxonomy

Kindom	**Animalia**
Phylum	**Chordata**
Sub-Phylum	**Vertebrata**
Class	**Amphibia**

- Amphibians are one of eight vertebrate classes
- From the Greek: *amphi* – both *bios* – life

Amphibian Biology

Two of the Major Amphibian Groups Inhabit North America

Caudata Salamanders

Ambystoma tigrinum

Anurans
Frogs/Toads

R. clamitans

Amphibian Biology (cont.)

- Ectotherms (cold-blooded)
  - Low metabolic rate
  - Moist permeable skin for oxygen exchange

R. pipiens

Amphibian Biology (cont.)

- Amphibian Breeding
  - Synchronized breeding
  - External fertilization
  - Biphasic life cycle

Amphibian Biology (cont.)

Habitat Use

- Adults/Juveniles
  - Terrestrial
  - Wetlands
- Egg/Larvae
  - Wetlands

Amphibian Biology (cont.)

Amphibian Trophic Status

- Predator
  - Algae and periphyton
  - Invertebrates
  - Small higher trophic organisms
- Prey
  - Predatory vertebrates
  - Adult invertebrates

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Phase 1: Literature Review and Development of Amphibian Screening Values

- Selection of Constituents of Potential Concern
- Literature Review
- Amphibian Screening Levels
  - Surface water toxicity
  - Sediment toxicity

R. catesbeiana

Constituents of Potential Concern

- 10 COPCs were selected for the detailed literature search because they are commonly identified at CERCLA, RCRA, and other investigated Navy sites

  - Cadmium
  - Chromium
  - Copper
  - Lead
  - Mercury
  - Nickel
  - Zinc
  - PCBs
  - 4,4 DDT
  - PAHs
  - OE Compounds

Literature Review Results

- Data limitations
  - Few data available and lack of standardized tests limits comparison of results
- Ecotoxicological data
  - Majority of ecotoxicological tests used surface water as exposure medium
  - Only two peer-reviewed studies with sediment or hydric soil were found

Amphibian Screening Levels

- Five of the 10 COPCs with most robust data set were evaluated further:
  - Four metals (Cd, Cu, Hg, and Zn)
  - One organochlorine (DDT)
  - Lethal effects data only (no sub-lethal)
  - Surface water data only
- Lethal effects percentile thresholds calculated
  - 10th percentile
  - 50th percentile
  - Evaluated relative to Ambient Water Quality Criteria (AWQC)

Recommendations

- Development of Standardized Toxicity Test
  - Early life stage North American species
  - Lethal and sub-lethal endpoints
- Validation of Test
  - High bioavailability spiked assays
  - Develop dose-response curve
  - Consider aging effects in hydric soil
  - Compounds to consider: divalent metals

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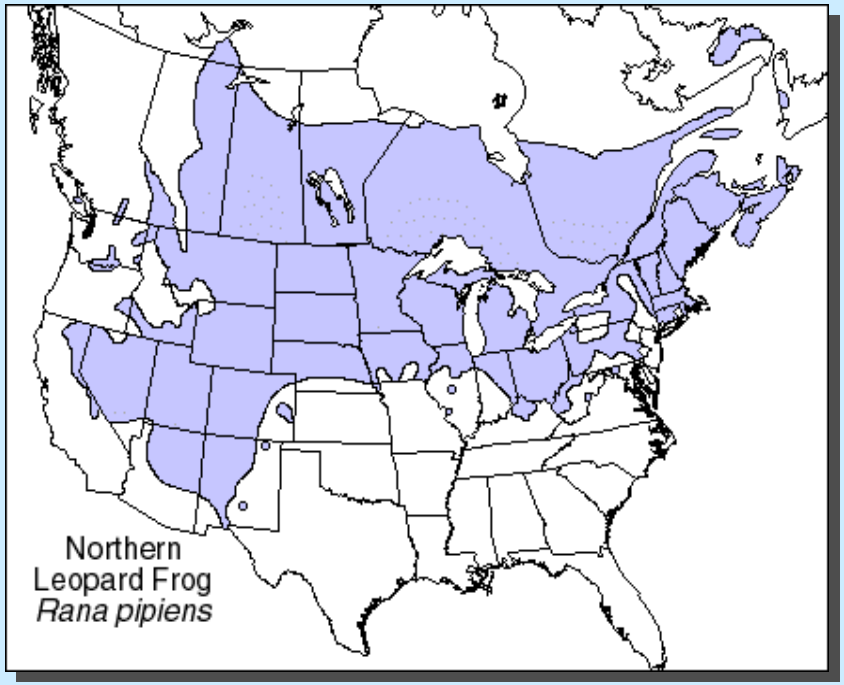
Phase 2: Development of Laboratory Testing Techniques for Amphibians Exposed to Sediment

- Short-Term Chronic Exposure Tests
  - Sediment exposure
  - Review of existing test methods
  - Produce draft standard operating procedure (SOP)

SOP Development

- Design a series of tests to identify:
  - Appropriate and available test species
  - Most sensitive age of test organisms
  - Appropriate test length
  - Appropriate test system
    - Flow-through or static test conditions
    - Food preference
    - Required volumes
  - Most sensitive test endpoints

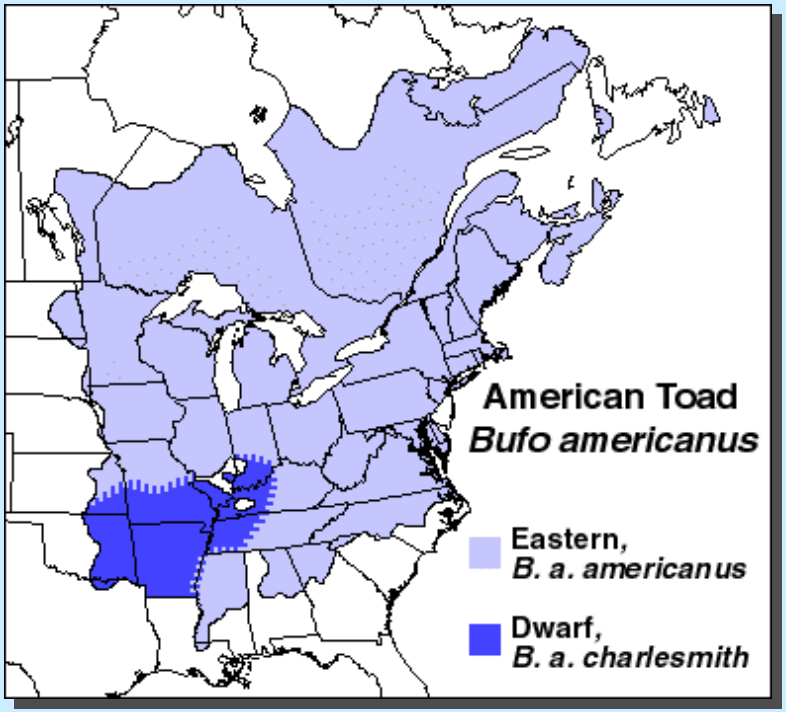
Northern Leopard Frog – *Rana pipiens*



- Small- to medium-sized
- Commercially available as tadpoles during breeding seasons

- Relatively short tadpole phase
- Documented developmental stages
- Native North American species
- Large habitat range

American Toad – *Bufo americanus*



- Small- to medium-sized
- May be commercially available as tadpoles
- Easily collected in the wild

- Relatively short tadpole phase
- Native North American species
- Wide habitat range covering much of eastern America

www.npwrc.usgs.gov/narcam/idguide/american.htm

General Observations

- Natural sediment serves as superior growth medium relative to artificial sediment
- Tetramin® is an acceptable food medium
- Flow-through tests recommended to eliminate ammonia buildup
- Growth (length and width) is best measurement to quantify sub-lethal effects on tadpoles
- Older tadpoles may be less sensitive to contaminants than younger tadpoles

Draft SOP

- 600 mL test vessels
- 100 mL sediment: 175 mL overlying water
- Recently hatched Rana or Bufo tadpoles
  - Before feeding starts
- Fed ground Tetramin® once feeding begins (Gosner stage 25)

Draft SOP (cont.)

- Eight replicates per treatment
- Five organisms per chamber (40 per treatment)
- Test duration of 10 days
  - Survival, body width, and body length
- Test temperature of 23±1°C
- Dissolved oxygen maintained above 3 mg/L
- Flow-through test system

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Phase 3: Validation of Testing Techniques

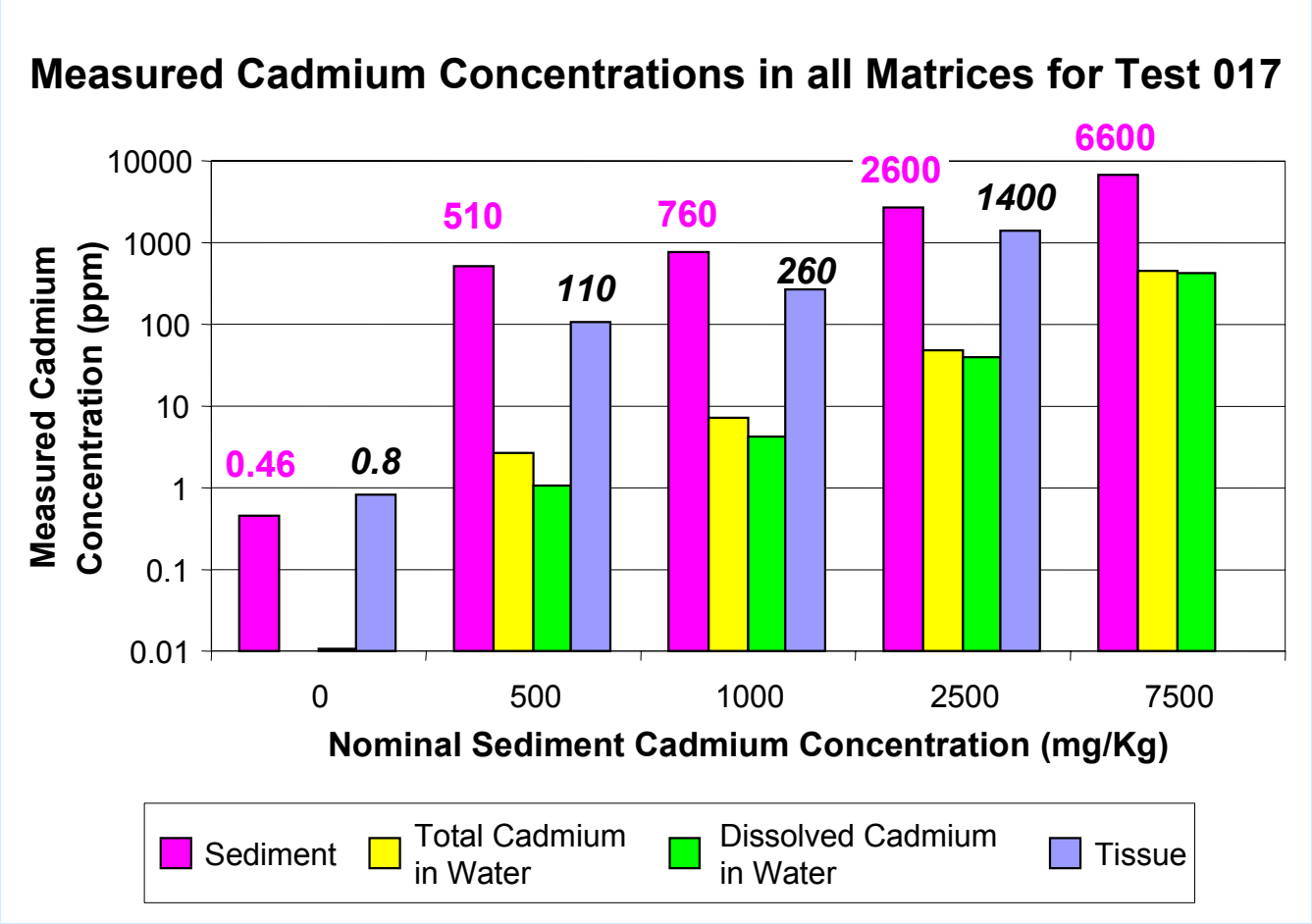
Confirmatory Phase

- Compare results with published literature
- Spiked sediment assays
  - Cadmium
  - Copper
  - Lead
  - Zinc
- Effects of organic carbon
  - Reduces bioavailability of

  Copper and Zinc

Validation Phase (cont.)

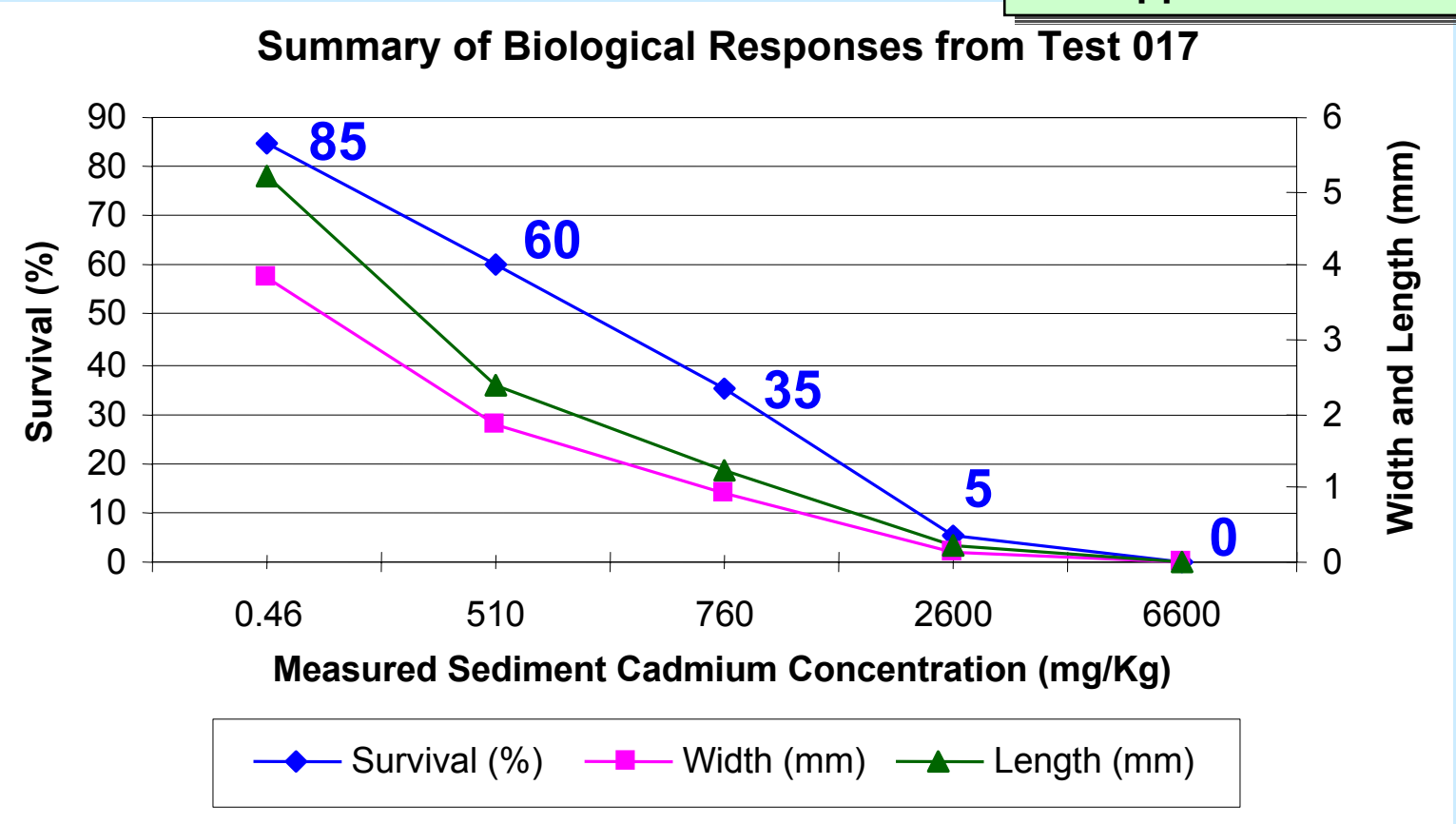
- Evaluated Cadmium Concentrations in All Matrices



Validation Phase (cont.)

- Evaluated Lethal and Sub-Lethal Endpoints

Survival NOEC
- 110 ppm in tissue
- 510 ppm in sediment

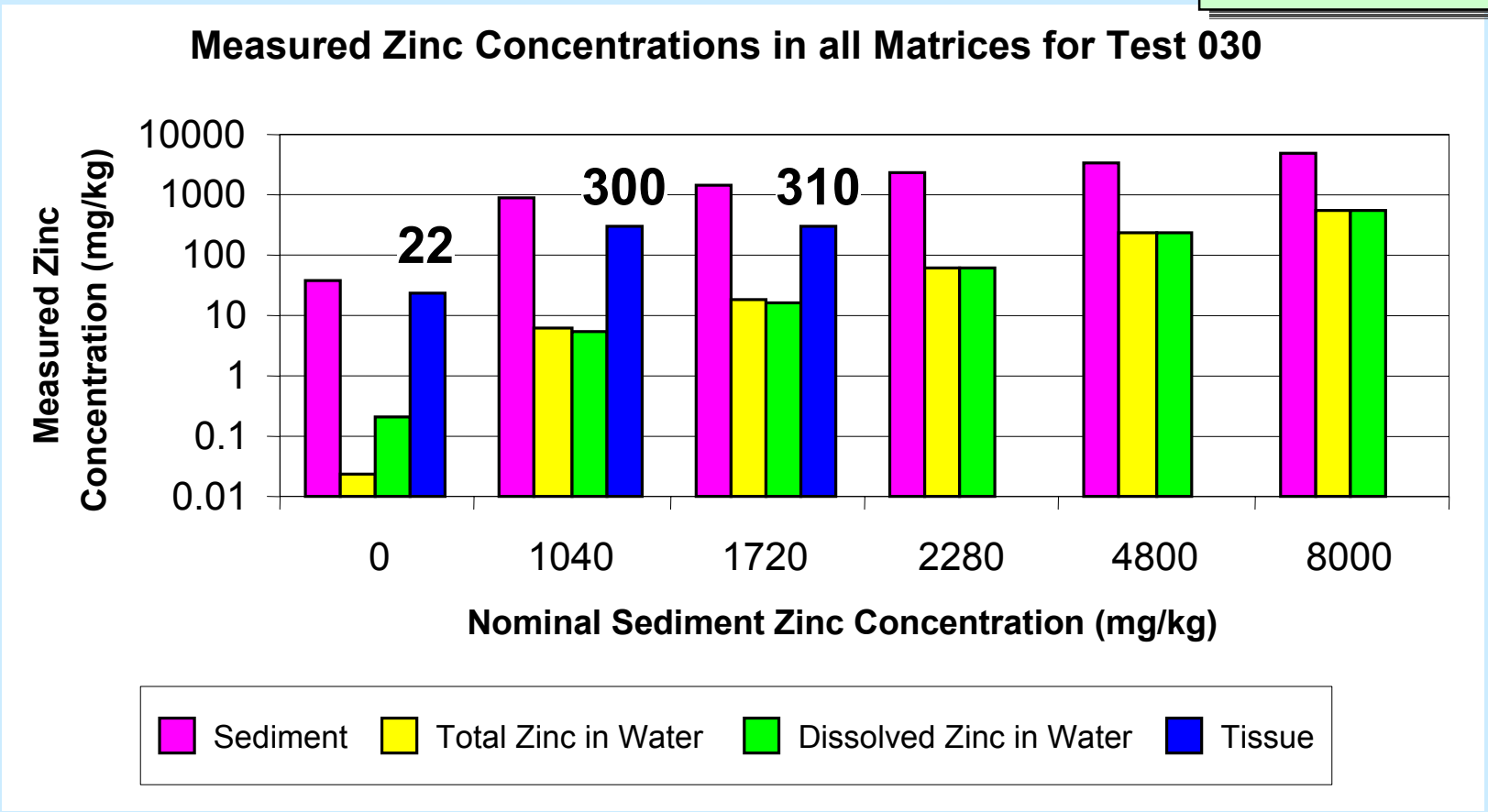


NOEC = No observed effects concentration

Validation Phase (cont.)

- Evaluated Zinc Concentrations in All Matrices

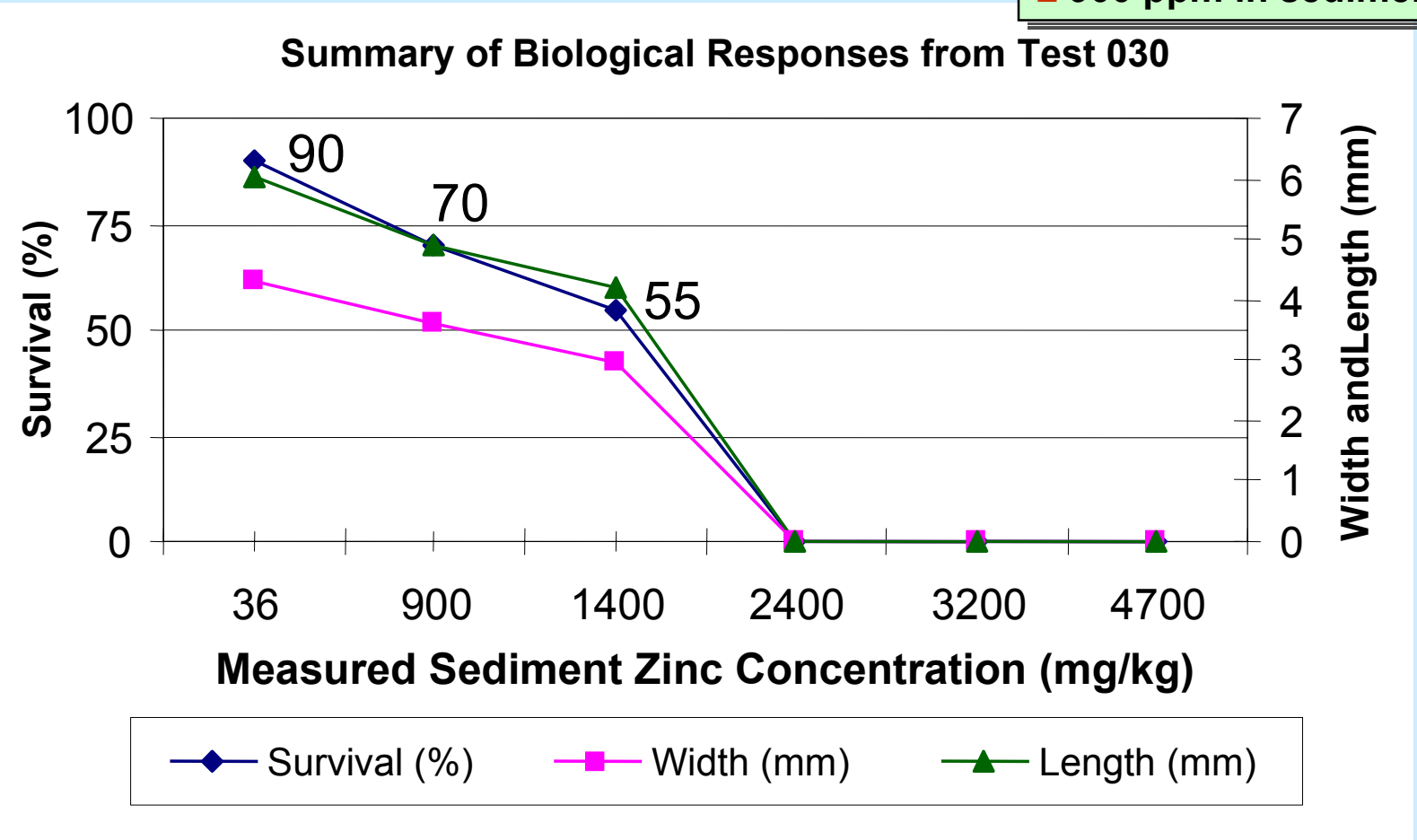
No survival in highest concentrations – no tissue at test termination



Validation Phase (cont.)

- Evaluated Lethal and Sub-lethal Endpoints

Survival NOEC
- 300 ppm in tissue
- 900 ppm in sediment



Validation Phase (cont.)

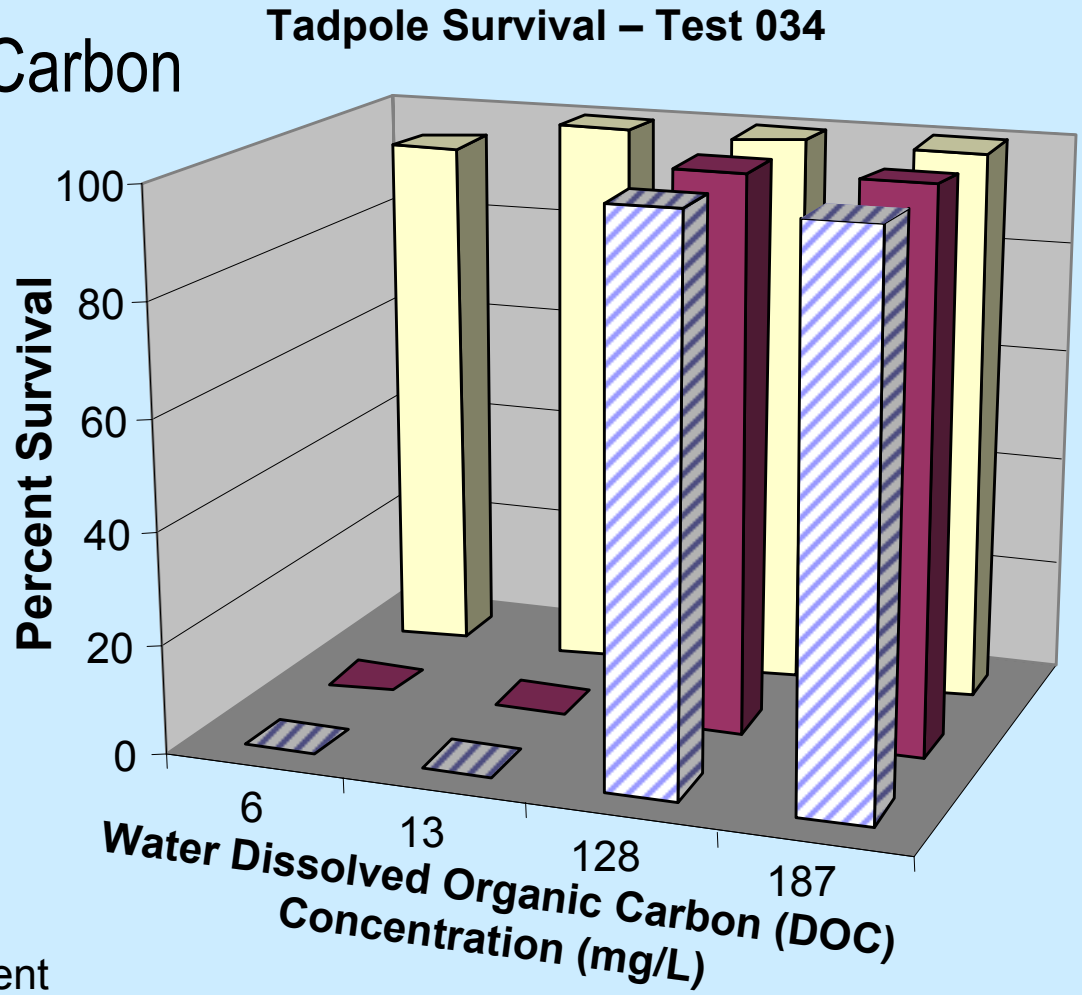
- Effects of Organic Carbon
  - Increased organic carbon resulted in decreased toxicity
    - Higher NOECs

Organic Carbon Concentration			Survival NOECs
Total in Sediment (mg/kg)	Total in Water (mg/L)	Dissolved in Water (mg/L)	Sediment Copper Concentration (mg/kg)
125	7	6	<1
1,300	32	13	5.2
13,000	155	128	250
14,000	223	187	420

Validation Phase (cont.)

- Effects of Organic Carbon

Increased organic carbon resulted in decreased toxicity



Validation Phase (cont.)

Results

- *Rana* & *Bufo* tadpoles are generally more tolerant of Cu, Cd, Pb, and Zn than test organisms used to develop sediment and water quality criteria

	Chronic AWQC* (µg/L)		Lowest IC_{25}** (µg/L)	
Surface Water	**Hardness 100 mg/L**	**Hardness 500 mg/L**	***Bufo***	***Rana***
Cadmium	0.25	0.84	1,000	540
* AWQC – Ambient Water Quality Criteria			** IC – Inhibition Concentration	
	Low Effect Levels (mg/kg)		**Lowest IC_{25}* (mg/kg)**	
Sediment	**LEL**	**ERL**	***Bufo***	***Rana***
Cadmium	0.6	1.2	540	230

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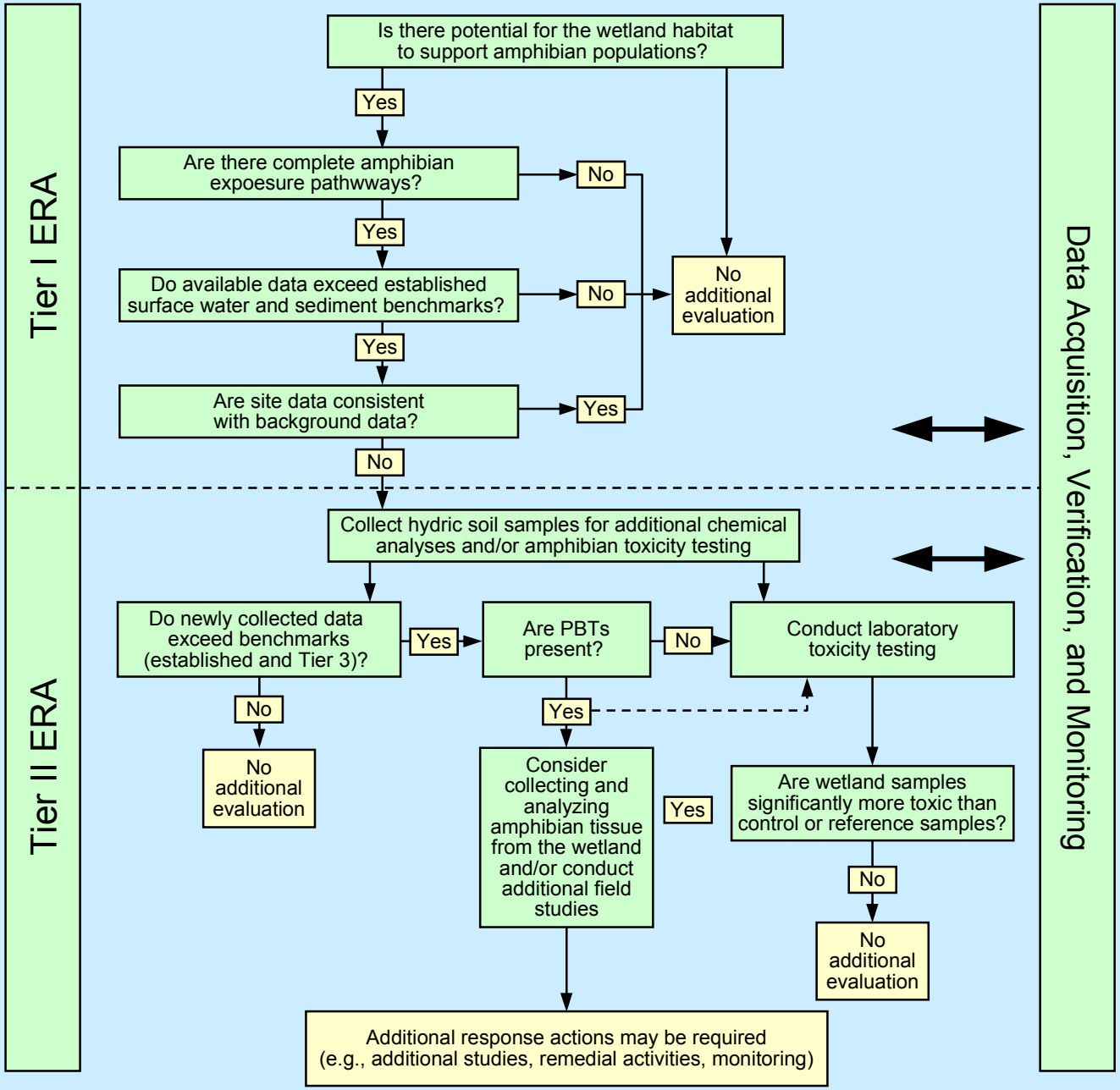
Phase 4:
Develop a Standardized Guidance Manual

- RPM guide describing how to assess risks to amphibians
- Appendices will include summary reports of Phases 1 through 3

R. pipiens

Preliminary RPM Guide Outline

Example Flow Chart to Assess Risk to Amphibians



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Cost Information

- 10-day sediment amphibian toxicity test is approximately $750 - $1,100 per sample
- Price range depends on number of samples – more samples allows a lower per-sample cost
- Standard *Hyalella azteca* or *Chironomus tentans* test is approximately $600 - $900 per sample

Summary and Conclusions

- Allow the Navy and DoD to develop more environmentally relevant risk assessments
- Risk managers can use this information to identify cleanup levels and set remediation goals
- Avoid costly and unnecessary wetland alteration based on use of inappropriate ecological endpoints
- Amphibian risk assessment web-based training tool will be available on NAVFAC's T2 website in May 2003

Questions